Electronic Supporting Information

Resolving the controls over the production and emission of ice-nucleating particles in sea spray

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Table S1. Diversity measures of bacteria in seawater and SSA in the *N. atomus* detritus addition experiment. In the SSA, days 1→2 and days 2→3 correspond to one day prior to, and peak INP emission days, respectively.

	2/13 Day 2 Water	2/12-13 Days 1→2 SSA	2/14 Day 3 Water	2/13-14 Days 2→3 SSA
Total OTUs	191	226	198	227
Chao 1	218	230	210	232
log series index (*α*)	16.8	20.5	17.9	20.8
Shannon index (*H*′)	2.27	2.41	2.34	3.01
Simpson’s index (*D*)	0.188	0.217	0.174	0.106

Table S2. Identities of dominant species in the *N. atomus* detritus addition experiment. Species with average relative abundances >0.1%, or >0.2% in any single sample, were included. For all these OTUs the closest isolates/sequences were from marine habitats. By far the most abundant bacterial species in the water was a *Marinobacterium* sp. (OTU 0). The type strain of this genus possesses a wide range of catabolic activities, utilizing various sugars, organic acids, amino acids, and other carbon sources.[1] OTU 2, a *Methylophaga* sp., increased greatly in relative abundance in the tank water between days 2 and 3 while maintaining a relatively low emission efficiency. While *Methylophaga* specializes in degrading C1 compounds, Sosa et al.[2] showed methylotrophs grew readily in seawater enriched with high molecular weight DOM from seawater, and Pinhassi et al. (2004)[3] found *Methylophaga marina* was associated with diatom-dominated microcosms; two *Methylophaga* spp. were recorded in the waveflume experiment studied by Michaud et al. (2018)[4], one of which comprised ~15% of the bacteria eight days after the start.

OTU1, a *Pseudofulvibacter*, was extraordinarily enriched in the aerosol while comprising only 0.01% of seawater sequences. *Pseudofulvibacter* belongs within the Flavobacteriia, a class that specializes in decomposing complex organic matter, especially polysaccharides and proteins, and can be enriched on particulate organic detritus and other surfaces.[5-7] Microorganisms associated with buoyant particles (e.g., microgels) that comprise the SML[8] contribute to the >10-fold enrichment of bacteria typically found in the SML.[9,10] Indeed, Aller et al. (2005)[9] observed that while bacteria were predominantly free-living in subsurface seawater, in lab-generated SSA ~60% were associated with particles. This concentration process may explain why the Flavobacteriia, in general, predominated among OTUs enriched ≥100-fold in the SSA (Fig. 4); Kuan (2015)[11] observed that *Flavobacteriaceae* accounted for most OTUs that could only be detected in the SML and not in the underlying water (i.e. they were completely partitioned at the surface).

Each incubation used a different starting batch of seawater (sampled at the same location). The initial bacterial diversity/evenness in each sample will have been strongly modified as a result of rapid growth of bacteria possessing the heterotrophic niche space/ability to exploit the substrate added. Viruses and nanoflagellates will, in turn, have been selected by the species of bacteria that proliferated. Due to this strong selection for rapidly growing heterotrophic bacteria, it’s reasonable to expect that the outcome of each incubation would be similar if replicated.

OTU	Avg. relative abund. (%)	Identity	Phylum (sub-phylum/Class)	Closest GenBank sequence	Similarity (%)
0	18.19	*Marinobacterium* sp.	Proteobacteria (Gamma)	*Marinobacterium*	98
1	17.78	*Pseudofulvibacter* sp.	Bacteroidetes (Flavobacteriia)	*Pseudofulvibacter* sp.	98
2	9.23	*Methylophaga* sp.	Proteobacteria (Gamma)	*Methylophaga* sp.	100
3	7.33	*Vibrio* sp.	Proteobacteria (Gamma)	*Vibrio* sp.	100
4	5.11	*Glaciecola* sp.	Proteobacteria (Gamma)	*Glaciecola* sp.	100
5	4.54	*Flavobactericeae* sp. 1	Bacteroidetes (Flavobacteriia)	*Flavobacteriaceae*	99
6	4.54	*Planktomarina* sp.	Proteobacteria (Alpha)	*Planktomarina temperata*	100
7	3.59	*Lentibacter* sp.	Proteobacteria (Alpha)	*Lentibacter algarum*	100
8	3.11	*Rhodobacteraceae* sp.	Proteobacteria (Alpha)	Unclassified *Rhodobacteraceae*	98
9	3.16	*Marinomonas* sp.	Proteobacteria (Gamma)	*Marinomonas blandensis*	100
10	2.27	*Arcobacter* sp.	Proteobacteria (Epsilon)	*Arcobacter lekithochrous*	100
11	2.15	*Flavobacteriaceae* sp. 2	Bacteroidetes (Flavobacteriia)	*Flavobacteriaceae*	97.3
13	1.22	*Lacinutrix* sp.	Bacteroidetes (Flavobacteriia)	*Lacinutrix*	98
14	1.17	*Pseudoalteromonas* sp.	Proteobacteria (Gamma)	*Pseudoalteromonas* sp.	100
15	1.17	*Colwellia* sp.	Proteobacteria (Gamma)	*Colwellia meonggei*	100
16	1.17	*Piscirickettsiaceae* sp.	Proteobacteria (Gamma)	*Piscirickettsiaceae* sp.	96
17	1.04	*Polaribacter* sp.	Bacteroidetes (Flavobacteriia)	*Polaribacter* sp.	98.9
18	1.13	*Neptuniibacter* sp.	Proteobacteria (Gamma)	*Neptuniibacter* sp.	99.7
19	0.90	*Colwellia* sp.	Proteobacteria (Gamma)	*Colwellia* sp.	99.2
20	0.79	*Cryomorphaceae* sp. 1	Bacteroidetes (Flavobacteriia)	Uncultured bacterium	98.6
21	0.75	*Formosa* sp.	Bacteroidetes (Flavobacteriia)	*Formosa* sp.	98.4
22	0.54	*Flavobactericeae* sp. 3	Bacteroidetes (Flavobacteriia)	Unclassified *Bacteroidetes*	99.7
23	0.45	*Flavobacteria* sp.	Bacteroidetes (Flavobacteriia)	*Flavobacteria* sp.	99.4
24	0.35	*Amylibacter* sp. 1	Proteobacteria (Alpha)	*Amylibacter* sp.	99.2
25	0.35	*Pelagibacter* sp.	Proteobacteria (Alpha)	*Pelagibacter* sp.	99.5
26	0.26	*Arcobacter* sp.	Proteobacteria (Epsilon)	*Arcobacter* sp.	100
27	0.21	*Rhodobacteraceae* sp.	Proteobacteria (Alpha)	*Rhodobacteraceae* sp.	97.9
28	0.25	*Psychrobium* sp.	Proteobacteria (Gamma)	*Psychrobium* sp.	98.9
29	0.20	*Flavobactericeae* sp. 4	Bacteroidetes (Flavobacteriia)	Uncultured *Flavobacteriaceae*	100
30	0.23	SAR86 clade sp. 1	Proteobacteria (Gamma)	Uncultured SAR86 bacterium	100
31	0.22	*Cobetia* sp.	Proteobacteria (Gamma)	*Cobetia* sp.	100
32	0.22	*Flavobacteriales* sp.	Bacteroidetes (Flavobacteriia)	Uncultured bacterium	100
33	0.21	*Amphritea japonica*	Proteobacteria (Gamma)	*Amphritea japonica*	100
34	0.22	*Piscirickettsiaceae* sp.	Proteobacteria (Gamma)	Uncultured marine bacterium (*Piscirickettsiaceae*)	100
35	0.18	*Halieaceae* sp.	Proteobacteria (Gamma)	Uncultured *Halieaceae*	100
37	0.17	*Phaeocystidibacter* sp.	Bacteroidetes (Flavobacteriia)	Uncultured *Flavobacteriia*	100
38	0.18	*Dokdonia* sp.	Bacteroidetes (Flavobacteriia)	*Dokdonia* sp.	99.7
39	0.16	*Oleibacter* sp.	Proteobacteria (Gamma)	*Oleibacter* sp.	99.4
40	0.14	*Flavobactericeae* sp. 5	Bacteroidetes (Flavobacteriia)	Uncultured *Flavobacteriaceae*	99.7
41	0.13	Gammaproteobacteria sp. 1	Proteobacteria (Gamma)	Uncultured Gammaproteobacteria	100
42	0.14	*Cryomorphaceae* sp. 2	Bacteroidetes (Flavobacteriia)	Uncultured *Cryomorphaceae*	99.2
43	0.11	*Amylibacter* sp. 2	Proteobacteria (Alpha)	*Amylibacter* sp.	97.6
44	0.11	Gammaproteobacteria sp. 2	Proteobacteria (Gamma)	Uncultured Gammaproteobacterium	99.7
45	0.11	*Flavobactericeae* sp. 6	Bacteroidetes (Flavobacteriia)	Uncultured *Flavobacteriaceae*	99.7
46	0.12	Alphaproteobacteria sp.	Proteobacteria (Alpha)	uncultured bacterium	100
47	0.11	*Luminiphilus* sp.	Proteobacteria (Gamma)	*Luminiphilus* sp.	97
48	0.11	*Antarctobacter* sp.	Proteobacteria (Alpha)	*Antarctobacter* sp.	99.2
50	0.10	SAR86 clade sp. 2	Proteobacteria (Gamma)	Uncultured SAR86 bacterium	100

51	0.11	*Alteromonas* sp.	Proteobacteria (Gamma)	*Alteromonas* sp.	98.9
52	0.10	*Enterovibrio* sp.	Proteobacteria (Gamma)	*Enterovibrio norvegicus*	100
53	0.09	*Alteromonadales* sp.	Proteobacteria (Gamma)	Uncultured *Alteromonadales* bacterium	97
54	0.08	*Aestuariicella* sp.	Proteobacteria (Gamma)	*Aestuariicella* sp.	98.9
55	0.08	Deltaproteobacteria sp.	Proteobacteria (Delta)	Unclassified Deltaproteobacteria	98.7
57	0.07	Gammaproteobacteria sp. 3	Proteobacteria (Gamma)	Uncultivated Gammaproteobacteria	100

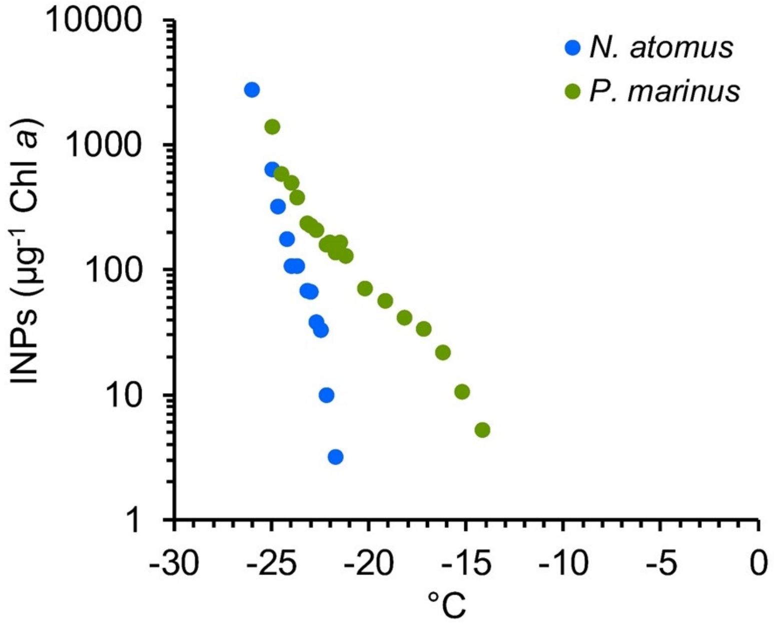

Fig. S1. Ice-nucleating particle concentrations of axenic cultures of *N. atomus* and *P. marinus* grown in f/2 and Pro99 media, respectively. Before conversion to INPs per chlorophyll equivalent units, results were corrected for INPs present in the media and temperatures of freezing adjusted by +2°C to account for freezing point depression of the media. The mass ratios of carbon to Chl *a* in the *N. atomus* and *P. marinus* cultures could be expected to be approximately 15[12].

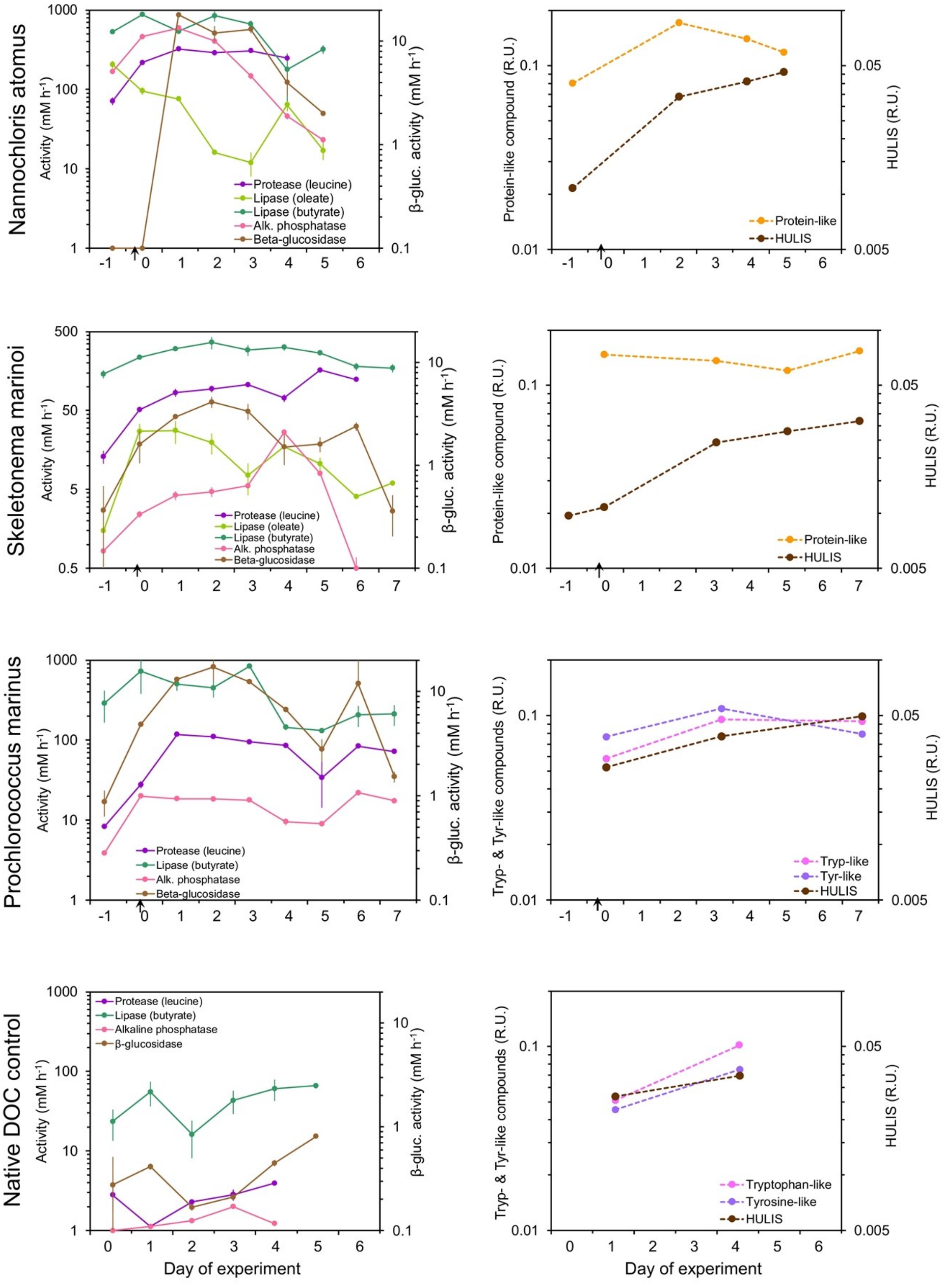

Fig. S2. Changes in degradative enzyme activities (left) and fluorescent molecules (right) in seawater during incubations, and in a no-addition control. Bars show ±SD.

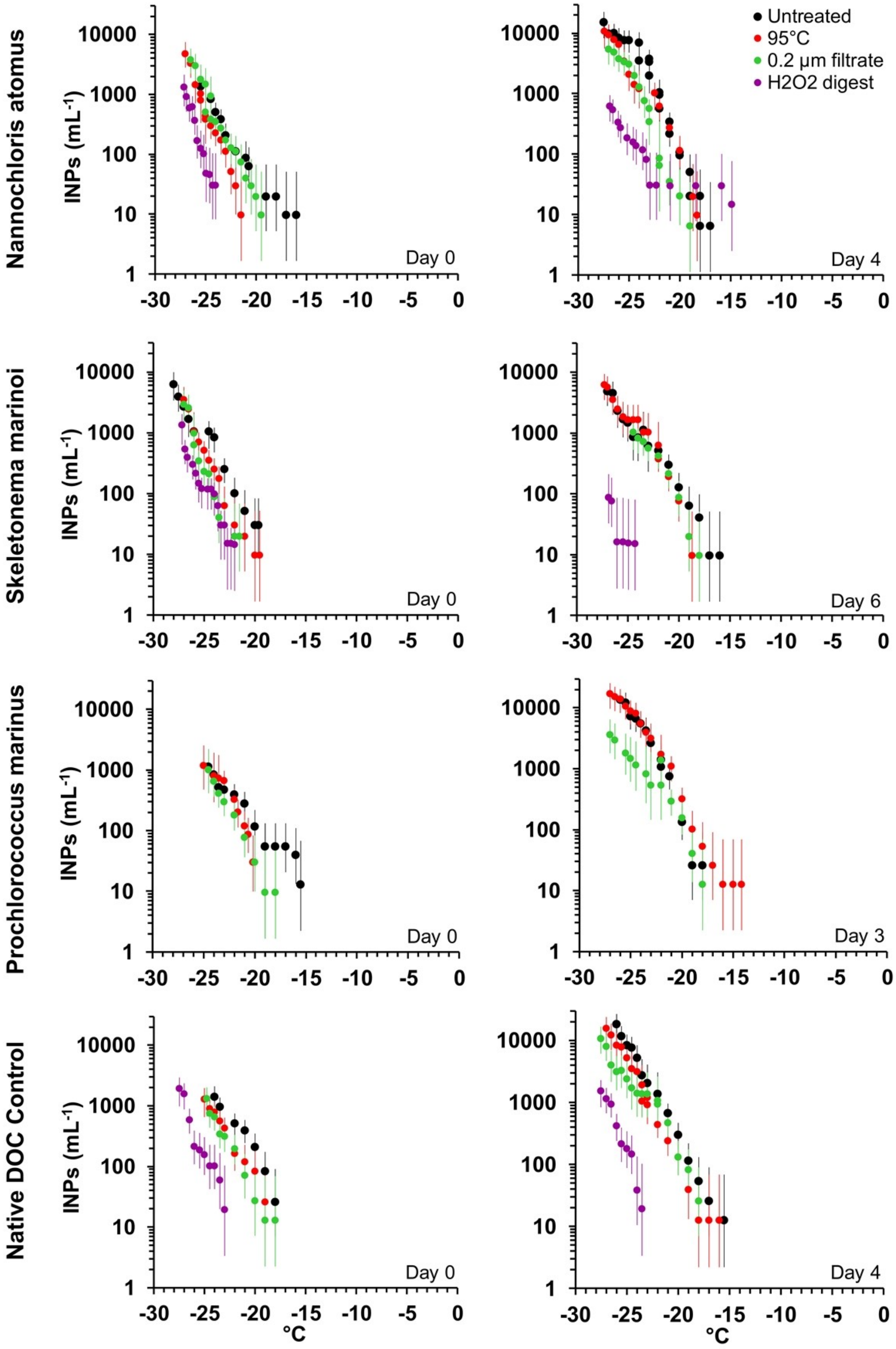

Fig. S3. Ice-nucleating entity abundance, size, sensitivity to being denatured by heat, and reductions caused by digestion with H_2O_2 in seawater at the start and at peak INE days during incubations, and in a no-addition control. Bars show 95% CIs.

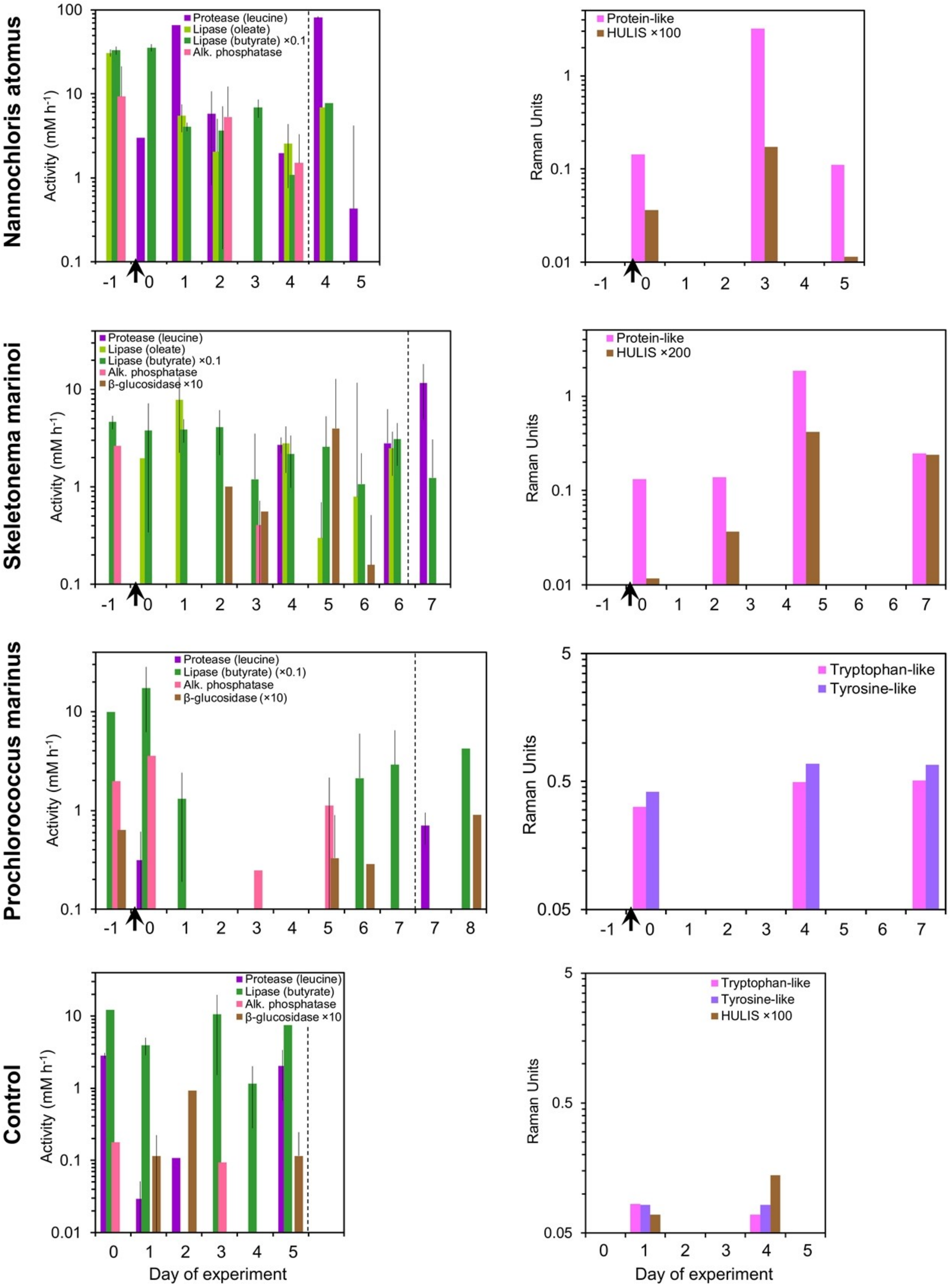

Fig. S4. Changes in degradative enzyme activities (left) and fluorescent molecules (right) in the SSA during incubations of seawater to which algal detritus had been added (at arrow), and in a no-addition control. Bars show ±SD. SSA EEM are scaled to per m^3 of air. No HULIS was measurable in the *P. marinus* experiment due to high Rayleigh scattering.

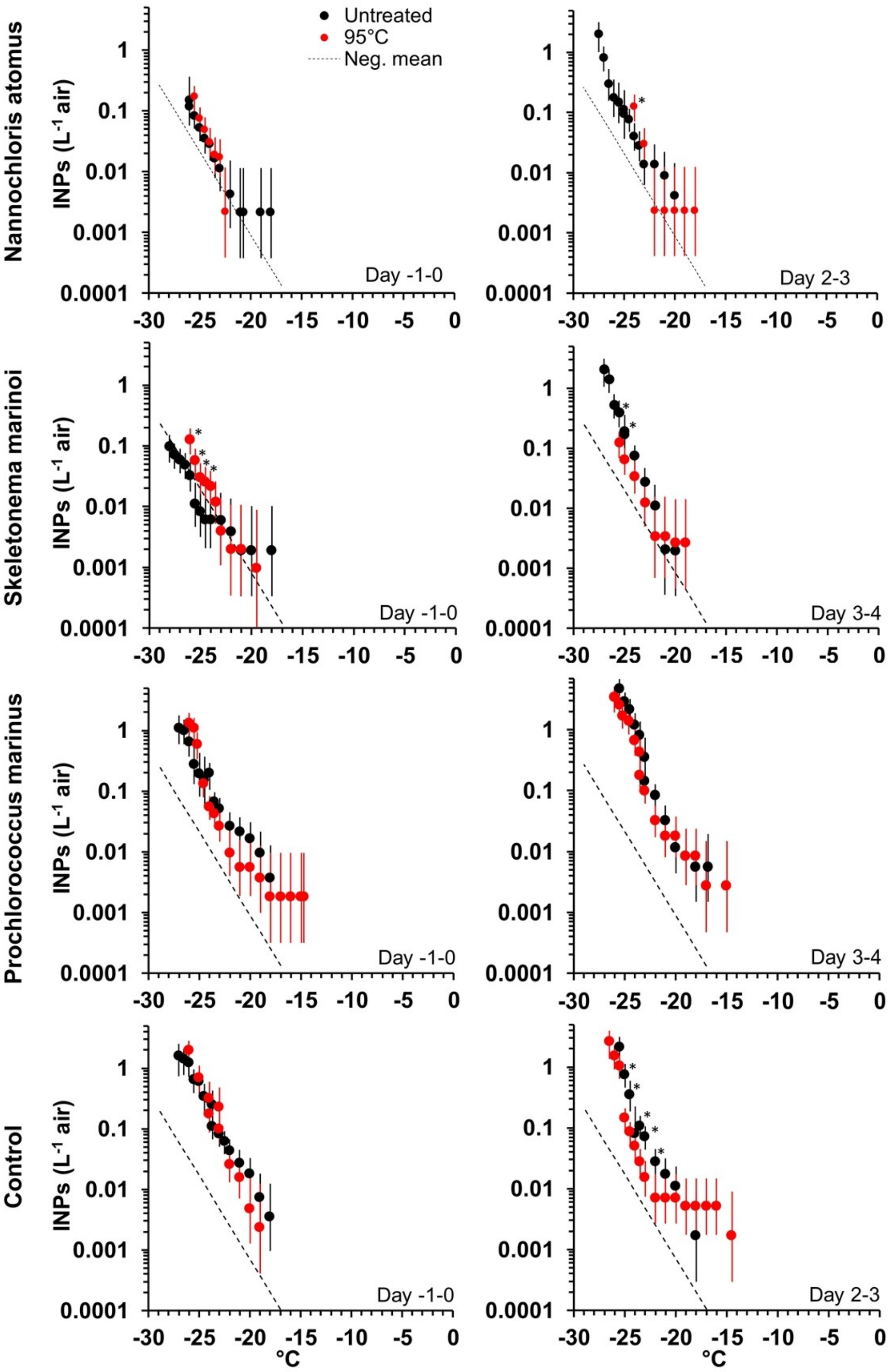

Fig. S5. Ice-nucleating particle abundance and sensitivity to being denatured by heat in SSA at the start and at peak INP emission days during incubations, and in a no-addition control. Bars show 95% CIs, and asterisks show significant differences ($p < 0.05$) using pairwise Fisher's Exact Tests. Dashed lines show results for the average of the blank filters when expressed on a per volume basis for each sample.

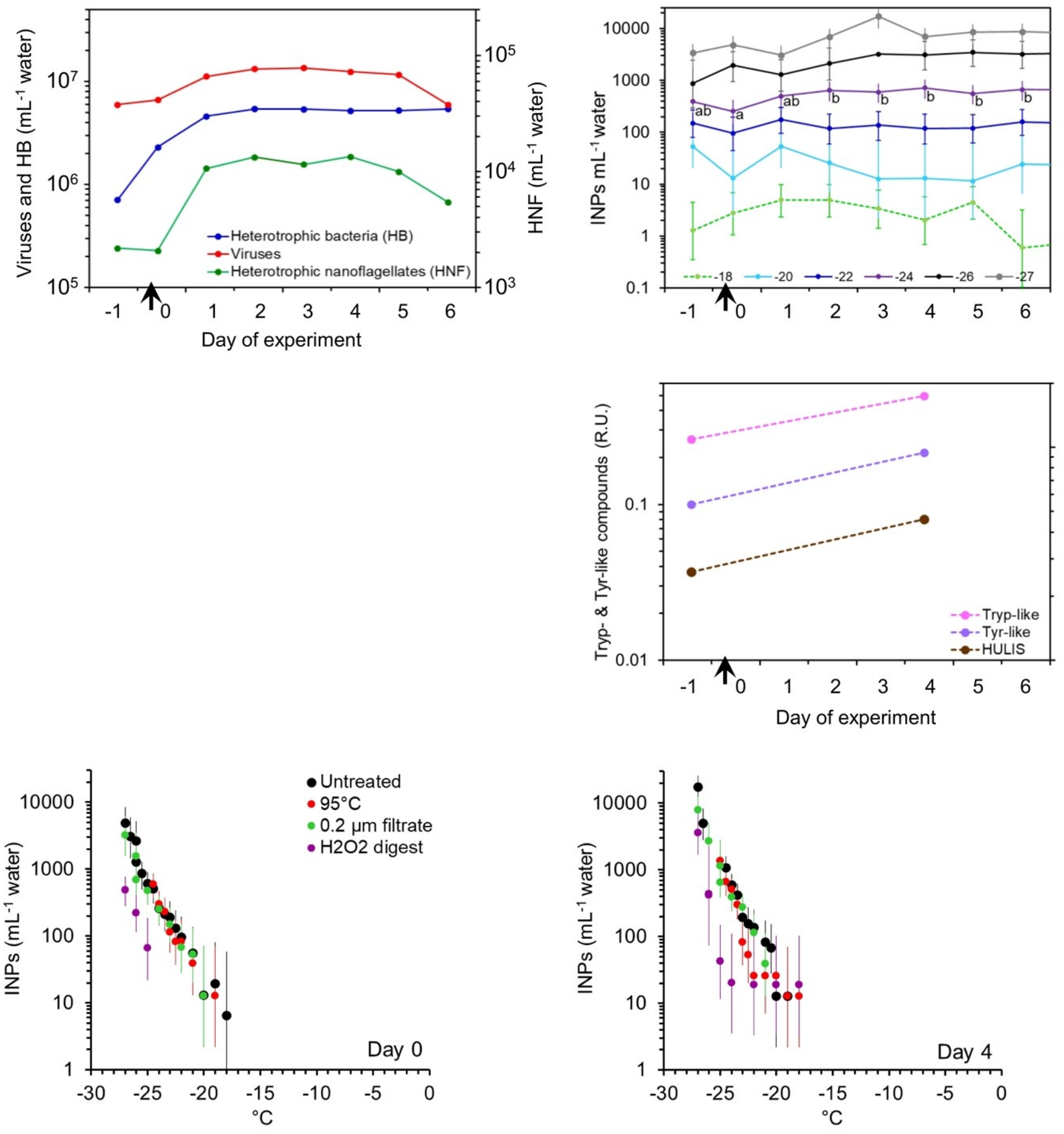

Fig. S6. Changes in microbial populations and INEs in filtered seawater to which BSA (1 mg L^{-1}) and glucose, galactose and mannose (each at 0.33 mg L^{-1}) had been added (arrow). For INE spectra, bars show 95% CIs and, for selected series, pairwise Fisher's exact tests were used to show the significance of changes (points sharing the same letter are not significantly different; *p* = 0.05). Also shown are fluorescent molecule abundances (centre), and, at bottom, INE size, and sensitivity to heat and H_2O_2 digestion at the start and peak INE days (bars show 95% CIs).

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